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		DB=PGPB,USPT; PLUR=YES; OP=OR		
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<u>L6</u>		L5 and (position ( probe and surface adj enhanced adj raman adj scattering adj spectroscopy and metallic surface and metallic coating ))	75	<u>L6</u>
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<u>L2</u>		Tsien.in.	127	<u>L2</u>
<u>L1</u>		6194548.pn.	1	<u>L1</u>

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## -continued

TTC GGC TAC GGC GTG CAG TGC TTC GCC CGC TAC CCC GAC CAC ATG AAG	240
Phe Gly Tyr Gly Val Gln Cys Phe Ala Arg Tyr Pro Asp His Met Lys	
305 310 315	
CAG CAG GAC TTC TTC AAG TCC GCC ATG CCC GAA GGC TAC GTC CAG GAG	288
Gln Gln Asp Phe Phe Lys Ser Ala Met Pro Glu Gly Tyr Val Gln Glu	
320 325 330	
CGC ACC ATC TTC TTC AAG GAC GAC GGC AAC TAC AAG ACC CGC GCC GAG	336
Arg Thr Ile Phe Phe Lys Asp Asp Gly Asn Tyr Lys Thr Arg Ala Glu	
335 340 345 350	
GTG AAG TTC GAG GGC GAC ACC CTG GTG AAC CGC ATC GAG CTG AAG GGC	384
Val Lys Phe Glu Gly Asp Thr Leu Val Asn Arg Ile Glu Leu Lys Gly	
355 360 365	
ATC GAC TTC AAG GAC GAC GGC AAC ATC CTG GGG CAC AAG CTG GAG TAC	432
Ile Asp Phe Lys Asp Asp Gly Asn Ile Leu Gly His Lys Leu Glu Tyr	
370 375 380	
AAC TAC AAC AGC CAC AAC GTC TAT ATC ATG GCC GAC AAG CAG AAG AAC	480
Asn Tyr Asn Ser His Asn Val Tyr Ile Met Ala Asp Lys Gln Lys Asn	
385 390 395	
GGC ATC AAG GTG AAC TTC AAG ATC CGC CAC AAC ATC GAG GAC GGC AGC	528
Gly Ile Lys Val Asn Phe Lys Ile Arg His Asn Ile Glu Asp Gly Ser	
400 405 410	
GTG CAG CCC GCC CAC TAC CAG CAG AAC ACC CCC ATC GGC GAC GGC	576
Val Gln Pro Ala Asp His Tyr Gln Gln Asn Thr Pro Ile Gly Asp Gly	
415 420 425 430	
CCC GTG CTG CTG CCC GAC AAC CAC TAC CTG AGC TAC CAG TCC GCC CTG	624
Pro Val Leu Leu Pro Asp Asn His Tyr Leu Ser Tyr Gln Ser Ala Leu	
435 440 445	
AGC AAA GAC CCC AAC GAG AAG CGC GAT CAC ATG GTC CTG CTG GAG TTC	672
Ser Lys Asp Pro Asn Glu Lys Arg Asp His Met Val Leu Glu Phe	
450 455 460	
GTG ACC GCC GCC GGG ATC ACT CAC GGC ATG GAC GAG CTG TAC AAG TAA	720
Val Thr Ala Ala Gly Ile Thr His Gly Met Asp Glu Leu Tyr Lys *	
465 470 475	

## (2) INFORMATION FOR SEQ ID NO: 4:

(i) SEQUENCE CHARACTERISTICS:

- (A) LENGTH: 239 amino acids
- (B) TYPE: amino acid
- (C) TOPOLOGY: linear

## (ii) MOLECULE TYPE: protein

## (xi) SEQUENCE DESCRIPTION: SEQ ID NO: 4:

Met Val Ser Lys Gly Glu Glu Leu Phe Thr Gly Val Val Pro Ile Leu  
 1 5 10 15

Val Glu Leu Asp Gly Asp Val Asn Gly His Lys Phe Ser Val Ser Gly  
 20 25 30

Glu Gly Glu Gly Asp Ala Thr Tyr Gly Lys Leu Thr Leu Lys Phe Ile  
 35 40 45

Cys Thr Thr Gly Lys Leu Pro Val Pro Trp Pro Thr Leu Val Thr <sup>Thr</sup>  
 50 55 60 <sup>64</sup>

<sup>Phe</sup> Gly Tyr Gly Val Gln Cys Phe Ala Arg Tyr Pro Asp His Met Lys  
 65 70 75 80

Gln Gln Asp Phe Phe Lys Ser Ala Met Pro Glu Gly Tyr Val Gln Glu  
 85 90 95

Arg Thr Ile Phe Phe Lys Asp Asp Gly Asn Tyr Lys Thr Arg Ala Glu  
 100 105 110

Val Lys Phe Glu Gly Asp Thr Leu Val Asn Arg Ile Glu Leu Lys Gly  
 115 120 125

-continued

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Ile	Asp	Phe	Lys	Asp	Asp	Gly	Asn	Ile	Leu	Gly	His	Lys	Leu	Glu	Tyr
130						135					140				
Asn	Tyr	Asn	Ser	His	Asn	Val	Tyr	Ile	Met	Ala	Asp	Lys	Gln	Lys	Asn
145						150					155				160
Gly	Ile	Lys	Val	Asn	Phe	Lys	Ile	Arg	His	Asn	Ile	Glu	Asp	Gly	Ser
	165						170					175			
Val	Gln	Pro	Ala	Asp	His	Tyr	Gln	Gln	Asn	Thr	Pro	Ile	Gly	Asp	Gly
	180						185					190			
Pro	Val	Leu	Leu	Pro	Asp	Asn	His	Tyr	Leu	Ser	Tyr	Gln	Ser	Ala	Leu
	195						200					205			
Ser	Lys	Asp	Pro	Asn	Glu	Lys	Arg	Asp	His	Met	Val	Leu	Leu	Glu	Phe
	210					215					220		221		
Val	Thr	Ala	Ala	Gly	Ile	Thr	His	Gly	Met	Asp	Glu	Leu	Tyr	Lys	
	225					230					235				

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What is claimed is:

1. A functional engineered fluorescent protein whose amino acid sequence is substantially identical to the amino acid sequence of Aequorea green fluorescent protein (SEQ ID NO:2) and which differs from SEQ ID NO:2 by at least the substitution T203X, wherein X is an aromatic amino acid selected from H, Y, W or F, and a substitution at S65, wherein the substitution is selected from S65G, S65T, S65A, S65L, S65C, S65V or S65I, and wherein said functional engineered fluorescent protein has a different fluorescent property than Aequorea green fluorescent protein.
2. The protein of claim 1, wherein the amino acid sequence differs from SEQ ID NO:2 by no more than the substitutions S65T/T203H; S65T/T203Y; S72A/F64L/S65G/T203Y; S72A/S65G/V68L/T203Y; S65G/V68L/Q69K/S72A/T203Y; S65G/S72A/T203Y; or S65G/S72A/T203W.
3. The protein of claim 1, wherein the amino acid sequence further comprises a substitution at Y66, wherein the substitution is selected from Y66H, Y66F, and Y66W.
4. The protein of claim 1, wherein the amino acid sequence further comprises a mutation at a position selected from the group consisting of Y145, N146, H148, M153, and V163.

5. The protein of claim 1, wherein the amino acid sequence further comprises a folding mutation selected from the group consisting of F64L, V68L and S72A.
6. The protein of any one of claims 1 to 5, which is a fusion protein wherein the fusion protein comprises a polypeptide of interest and the functional engineered fluorescent protein.
7. The protein of claim 1, wherein the amino acid sequence differs from SEQ ID NO:2 by no more than the substitutions S65T/T203H; S65T/T203Y; S72A/F64L/S65G/T203Y; S72A/S65G/V68L/T203Y; S65G/V68L/Q69K/S72A/T203Y; or S65G/S72A/T203Y.
8. The protein of claim 1, wherein the amino acid sequence differs from SEQ ID NO:2 by no more than the substitutions S65G/S72A/K79R/T203Y.
9. The protein of claim 1, wherein the amino acid sequence differs from SEQ ID NO:2 by no more than the substitutions T203Y/S65G/V68L/S72A.

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